BIOPHARMACEUTICS & DRUG DISPOSITION
Biopharm. Drug Dispos. **29**: 382–395 (2008)
Published online 11 June 2008 in Wiley InterScience
(www.interscience.wiley.com) **DOI**: 10.1002/bdd.622

Pharmacokinetics of Budesonide and Formoterol Administered via a Series of Single-Drug and Combination Inhalers: Four Open-Label, Randomized, Crossover Studies in Healthy Adults

Annika Eklund[a], Ann Tronde[a,*], Ingegerd Johannes-Hellberg[a], Michael Gillen[b] and Lars Borgström[a]
[a] *AstraZeneca, Lund, Sweden*
[b] *AstraZeneca, Wilmington, DE, USA*

ABSTRACT: *Objective*: To investigate the pharmacokinetics of budesonide and formoterol administered concomitantly in healthy adults.

Methods: Three single-dose, open-label crossover studies (n=28 each) were conducted (Study I: budesonide pMDI, formoterol DPI, budesonide pMDI+formoterol DPI; Study II: budesonide/formoterol pMDI, budesonide pMDI+formoterol DPI; Study III: budesonide/formoterol pMDI [three budesonide formulation strengths; constant formoterol]). Study IV (n=28) assessed steady state pharmacokinetics (budesonide/formoterol pMDI [two/four inhalations twice daily, 5-day treatment; four inhalations, single-dose]).

Results: Study I: no pharmacokinetic interactions were observed between budesonide and formoterol. Study II: *AUC* ratios were 97.9% (budesonide) and 82.2% (formoterol) (budesonide/formoterol pMDI versus budesonide pMDI+formoterol DPI). Study III: formoterol *AUC* was comparable across budesonide/formoterol pMDI formulation strengths; budesonide *AUC* increased with formulation strength in proportion to fine particle dose. Study IV: dose proportionality was demonstrated for budesonide (*AUC* ratio, 104.3%) and suggested for formoterol (*AUC* ratio, 117.6%) with budesonide/formoterol pMDI (steady state); budesonide and formoterol *AUC* was higher with repeated versus single-dose budesonide/formoterol pMDI (four inhalations).

Conclusions: No pharmacokinetic interactions were observed between budesonide and formoterol. Budesonide dose variation in budesonide/formoterol pMDI did not affect formoterol exposure. Steady state budesonide/formoterol pMDI dose-doubling yielded proportional increases in budesonide and formoterol exposure.

Key words: pharmacokinetics; bioavailability; budesonide; formoterol; Symbicort; budesonide/formoterol pMDI; combination inhaler

Introduction

Inhaled corticosteroids (ICSs) are the recommended first-line treatment for persistent asthma, while the addition of a long-acting β_2-adrenergic agonist (LABA) to ICS therapy is preferred for patients whose asthma is not controlled on a medium dose of ICS alone [1,2]. This combination also is appropriate for the treatment of patients with symptomatic chronic obstructive pulmonary disease (COPD) who have a postbronchodilator forced expiratory volume in 1 s (FEV_1) < 50%

*Correspondence to: AstraZeneca R&D Lund, SE-221 87 Lund, Sweden.
Email: Ann.Tronde@astrazeneca.com

Received 28 June 2007
Revised 27 March 2008
Accepted 27 March 2008

predicted and a history of repeated exacerbations [3]. The ICS budesonide and the LABA formoterol are available in combination in a dry powder inhaler (DPI) (Symbicort® Turbuhaler®, AstraZeneca, Lund, Sweden) and in a pressurized metered-dose inhaler (pMDI) (Symbicort® Inhalation Aerosol, AstraZeneca, Wilmington, DE, USA). Combining these two controller medications in one inhaler simplifies the dosing regimen for patients for whom combination therapy is appropriate, which may be beneficial, as the use of multiple medications may be confusing for patients and may negatively affect their adherence to a prescribed treatment [4,5]. In addition, the use of a combination inhaler incorporating both ICS and LABA in patients with asthma ensures that the LABA is not administered alone [2].

Randomized, double-blind clinical trials have demonstrated the safety and efficacy of budesonide/formoterol DPI in maintaining lung function and controlling symptoms in adult and pediatric patients with persistent asthma and that combination therapy is more efficacious than budesonide alone [6–10]. In patients with COPD, the same combination prolonged the time to first exacerbation compared with either treatment alone or placebo [11] and reduced the number of severe exacerbations [12]. The pharmacokinetic profiles of budesonide and formoterol have been reported when each agent is administered separately [13–19] and combined via a DPI [20]. Importantly, systemic exposures to budesonide and formoterol were comparable after administration together in the DPI compared with administration in separate inhalers [20]. Concomitant dosing did not result in any pharmacokinetic interactions [20].

The aim of the present investigation was to evaluate the pharmacokinetics and systemic bioavailability of budesonide and formoterol administered via the recently introduced hydrofluoroalkane (HFA) budesonide/formoterol pMDI. Two of the studies (I and II) included assessments of budesonide and formoterol pharmacokinetics when delivered by a single drug inhaler. In these studies, budesonide was delivered from an HFA pMDI formulation that was identical to budesonide/formoterol pMDI with the exception of the inclusion of formoterol. This formulation was produced specifically for use in clinical trials and is not a marketed product. Formoterol was administered as Oxis® Turbuhaler® (AstraZeneca, Lund, Sweden), which is the dry powder inhaler formulation available in many countries. These products were included in these pharmacokinetic studies because they were the monoproducts used as comparators in the phase III program for budesonide/formoterol pMDI to fulfil regulatory requirements for the evaluation of a combination product.

By comparing the pharmacokinetics of budesonide and formoterol when delivered separately or in combination, it may be possible to detect any alterations in systemic exposure due to changes in formulation.

Methods

Study designs and subjects

Four randomized crossover studies were conducted (SD-039-0722 [Study I], SD-039-0721 [Study II], SD-039-0723 [Study III], and SD-039-0724 [Study IV]). All treatments were administered in an open-label manner; subjects were trained to ensure correct inhalation technique for each device, and each inhalation was supervised by a study nurse. In each study, the time between each inhalation was such that the total inhalation times were similar for each treatment: the time between each inhalation was extended for the treatments administered via one inhaler to equal the total inhalation times of the treatments administered via separate inhalers. No unnecessary pauses were allowed between inhalations for treatments administered via separate inhalers. The maximum total inhalation time ranged from 4 to 7 min in the four studies. Inhalations and blood and urine sampling were conducted at the AstraZeneca Clinical Pharmacology Unit (Lund, Sweden).

Treatments for each study are shown in Table 1. In each study, the subjects received the treatments in random order. Study I investigated the pharmacokinetic interaction of budesonide and formoterol when administered simultaneously as budesonide pMDI+formoterol DPI (concomitant administration) compared with each product administered alone. Study II was

DOI: 10.1002/bdd

Table 1. Study treatments

Study	Treatment regimen (total delivered dose)	Product/trade name[a]
I	Eight inhalations:	
	BUD pMDI 160 μg (1280 μg)	Not marketed
	FM DPI 4.5 μg (36 μg)	Oxis® Turbuhaler®
	BUD pMDI 160 μg (1280 μg)+FM DPI 4.5 μg (36 μg)	Not marketed+Oxis® Turbuhaler®
	3–14 day washout period between treatments	
II	Eight inhalations:	
	BUD/FM pMDI 160/4.5 μg (1280/36 μg)	Symbicort® pMDI
	BUD pMDI 160 μg (1280 μg)+FM DPI 4.5 μg (36 μg)	Not marketed+Oxis® Turbuhaler®
	3–14 day washout period between treatments	
III	Twelve inhalations:	
	BUD/FM pMDI 40/4.5 μg (480/54 μg)	Not marketed
	BUD/FM pMDI 80/4.5 μg (960/54 μg)	Symbicort® pMDI
	BUD/FM pMDI 160/4.5 μg (1920/54 μg)	Symbicort® pMDI
	5–14 day washout period between treatments	
IV	Two inhalations b.i.d. × 5 days:	
	BUD/FM pMDI 160/4.5 μg (320/9 μg per day)	Symbicort® pMDI
	Four inhalations b.i.d. × 5 days:	
	BUD/FM pMDI 160/4.5 μg (640/18 μg per day)	Symbicort® pMDI
	Four inhalations single dose:	
	BUD/FM pMDI 160/4.5 μg (640/18 μg)	Symbicort® pMDI
	5–28 day washout period between treatments	

BUD, budesonide; pMDI, pressurized metered-dose inhaler; FM, formoterol; DPI, dry powder inhaler; b.i.d. twice daily.
[a]Symbicort® pMDI is manufactured by AstraZeneca, Wilmington, DE, USA; Oxis® Turbuhaler® is manufactured by AstraZeneca, Lund, Sweden. The budesonide pMDI used in these studies was designed as a comparator and is not marketed in any country.

designed to investigate the relative systemic bioavailability of budesonide and formoterol when inhaled from budesonide/formoterol pMDI (one inhaler) compared with budesonide pMDI+formoterol DPI (concomitant administration) and to compare plasma and urine sampling when estimating the relative systemic bioavailability of formoterol. Study III investigated the dose proportionality of budesonide, measured as systemic bioavailability, and the relative systemic bioavailability of formoterol when administered via three different formulation strengths of budesonide/formoterol pMDI. Study IV was designed to investigate (i) the dose proportionality of budesonide and formoterol, measured as systemic bioavailability, when inhaled from budesonide/formoterol pMDI at two different doses twice daily for 5 days and (ii) the relationship between single- and repeated-dose pharmacokinetics of budesonide and formoterol when inhaled from budesonide/formoterol pMDI.

All study protocols were approved by the Independent Ethics Committee in Lund, Sweden, conducted in accordance with the ethical principles of the Declaration of Helsinki and applicable local regulations, and consistent with Good Clinical Practice. Written informed consent was obtained before any study procedures were initiated.

For all studies, healthy subjects aged 18 to 55 years with a body mass index between 18 and 30 kg/m^2 were included. Women of childbearing age were to use reliable contraception, and no pregnant or lactating women were included in the studies. Subjects using any regular medication or therapy including oral contraceptives, over-the-counter remedies, herbal preparations, vitamins and mineral supplements were excluded. Subjects with an acute illness or intake of prescribed medication within 2 weeks before enrolment; history or evidence of any significant disease or disorder; or a known or suspected hypersensitivity to corticosteroids, β_2-adrenergic agents, inhaled lactose or other excipients in the study drugs also were excluded. Subjects could not participate in another study within 3 months before screening, apart from noninvasive methodology studies in which no drugs were given.

The following restrictions were associated with participation in the studies to standardize condi-

DOI: 10.1002/bdd

Table 2. Baseline and demographic characteristics[a] of subjects in each study

	Study I (n=28)	Study II (n=28)	Study III (n=28)	Study IV (n=28)
Age (y)	26 (6)	24 (3)	25 (5)	26 (4)
Range	19–47	20–29	19–34	21–36
Weight (kg)	69 (12)	71 (12)	72 (12)	72 (11)
Range	51–90	54–106	54–99	58–96
Height (cm)	174 (9)	177 (9)	176 (9)	175 (7)
Range	161–192	163–191	156–195	160–190
BMI (kg/m^2)	23 (3)	23 (2)	23 (2)	23 (2)
Range	18–29	20–29	19–29	20–28

[a]All values are presented as mean (standard deviation) unless otherwise indicated.

tions and to limit the risk of drug–drug and drug–food interactions, as well as to ensure subject safety and welfare. Subjects were required to abstain from taking any prescribed medication and any nicotine-containing products during the study; refrain from strenuous physical activity and drinking alcohol from 24 h before and during all visits; and abstain from intake of any nonprescribed medication (except paracetamol if needed), grapefruit and grapefruit juice from 72 h before and during all treatment periods. Subjects were required to fast (water allowed) for at least 10 h at the time of arrival at the Clinical Pharmacology Unit in the morning during the treatment periods (in the morning of the last treatment day for the repeated-dose treatments in Study IV) and for 3 h at the time of arrival at enrolment and follow-up. Subjects were served a standardized breakfast, which had to be finished 30 min before treatment. No food or liquids were allowed until 4 h after treatment, except for water, which was allowed 1 h after treatment.

Sample collection

Blood samples for analysis of budesonide and formoterol were collected predose (0 min) in all studies using an indwelling plastic cannula inserted into the forearm vein. In all studies, postdose samples were collected 10, 20, 40 and 60 min, and 2, 4, 6, 8, 10 and 12 h after treatment with study drug. In Study III, postdose samples were also collected at 14, 16, 18, 20, 22 and 24 h after treatment with the study drug for determination of formoterol pharmacokinetics. In Study IV, for the repeated-dose treatments, postdose samples were collected on the last treatment day. In Study II, all urine was collected over the 48 h postdose period for the determination of total excreted formoterol.

In all studies, blood samples were collected into sodium heparinized tubes at each designated time. The samples were centrifuged at $1500 \times g$ for 10 min at room temperature. For budesonide, the plasma was then transferred to cryotubes and stored below $-20^{\circ}C$ before analysis. For formoterol, plasma was immediately transferred to cryotubes containing citric acid and stored below $-20^{\circ}C$ before analysis. Urine samples were stored below $-20^{\circ}C$.

Formoterol

Quantitative determination of formoterol base (MW: 344.4 g/mole) in plasma and urine was performed by Quintiles AB Analytical Services (Uppsala, Sweden). The plasma and urine samples were prepared by adding a $^2H_4$-labeled analogue of formoterol as an internal standard followed by solid phase extraction (Isolute CBA, Sorbent AB, Sweden). Analysis was performed by coupled column reversed-phase liquid chromatography with electrospray tandem mass spectrometric detection (LC-ESI-MS/MS) (Instrument: Quattro II with Z-spray, Waters Corporation, Micromass UK Ltd, Manchester, UK). Chromatography was performed using a gradient of water, methanol and acetic acid on a coupled column LC system comprising a CN-window column, a C_{18}-collection column and a C_{18}-analytical column (ACE C_{18} 50×2.1 mm i.d., Hichrome Ltd, UK). Formoterol and the internal standard were detected using ESI positive ion

DOI: 10.1002/bdd

Table 3. Comparisons of pharmacokinetic parameters[a] for budesonide pMDI and formoterol DPI (study 1)

Parameter	BUD pMDI 8×160 µg	FM DPI 8×4.5 µg	BUD pMDI 8×160 µg +FM DPI 8×4.5 µg	Treatment comparisons, mean ratio (90% CI)	
				BUD pMDI[b]+FM DPI[c] vs BUD pMDI[b]	BUD pMDI[b]+FM DPI[c] vs FM DPI[c]
Budesonide					
AUC, nmol.h/l	13.9 (13.2–14.6)	—	13.0 (12.4–13.7)	93.8 (87.5–100.6)	—
C_{max}, nmol/l	3.8 (3.5–4.1)	—	4.0 (3.6–4.3)	105.1 (93.6–117.9)	—
$t_{1/2}$, h	3.4 (3.2–3.7)	—	3.6 (3.3–3.8)	104.3 (95.1–114.4)	—
Formoterol					
AUC, pmol.h/l	—	587.3 (556.1–620.3)	592.4 (561.0–625.6)	—	100.9 (93.4–109.0)
C_{max}, pmol/l	—	180.5 (170.7–190.9)	178.9 (169.2–189.1)	—	99.1 (91.6–107.2)
$t_{1/2}$, h	—	5.9 (5.2–6.6)	6.5 (5.8–7.2)	—	109.6 (93.0–129.1)

pMDI, pressurized metered-dose inhaler; DPI, dry powder inhaler; CI, confidence interval; BUD, budesonide; FM, formoterol; *AUC*, area under the plasma concentration versus time curve from time zero to infinity; C_{max}, maximum plasma concentration; $t_{1/2}$, elimination half-life.

[a]Values represent geometric means for all parameters; comparisons are shown as percentages based on ratios (90% confidence interval).

[b]Eight inhalations of BUD pMDI 160 µg.

[c]Eight inhalations of FM DPI 4.5 µg.

multiple reaction monitoring (MRM) of the transitions *m/z*: 345.00–149.20 (formoterol) and 349.00–153.30 (internal standard).

The plasma method was validated over the concentration range 5.00 to 1000 pmol/l with a lower limit of quantification (LLOQ) of 5.00 pmol/l. The inter-assay repeatability was 3.0 to 6.6%, and the accuracy was better than 8.4%. The urine method was validated over the concentration range 40.0 to 50000 pmol/l with a LLOQ of 40.0 pmol/l using a 1.0 ml sample volume. The inter-assay repeatability was 5.0 to 11.1%, and the accuracy was better than 4.6%.

Budesonide

Quantitative determination of R/S budesonide (MW: 430.24 g/mole) in plasma was performed by TNO Nutrition and Food Research (Zeist, the Netherlands). The plasma samples were prepared by adding a $^2H_8$-labeled analogue of budesonide as an internal standard followed by solid phase extraction (Isolute MF C18, Sorbent AB, Sweden). Analysis was performed by liquid chromatography–atmospheric pressure chemical ionization–tandem mass spectrometry (LC-APCI-MS/MS) (Instrument: Finnigan TSQ 7000, ThermoFinnigan, San José, CA, USA). Chromatography was performed using a gradient of water, methanol and acetic acid on an LC system comprising a C8-guard column and a C8-analytical column (Zorbax SB-C8 30×4.6 mm i.d., Agilent Technologies, Sweden). Budesonide and the internal standard were detected using negative ion multiple reaction monitoring (MRM) of the transitions of the acetate adduct of budesonide (*m/z*: 489–357) and its internal standard (*m/z*: 497–357). The plasma method was validated over the concentration range 0.010 to 10 nmol/l with a LLOQ of 0.01 nmol/l. The inter-assay repeatability was 2.4 to 13.0%, and the accuracy was within the range of to −4.0 to 0.2%.

Calculation of pharmacokinetic variables

The plasma concentration data for budesonide and formoterol were described by standard pharmacokinetic variables: area under the curve of plasma concentration versus time (*AUC*) and maximum plasma concentration (C_{max}). Values for each subject were estimated with standard

DOI: 10.1002/bdd

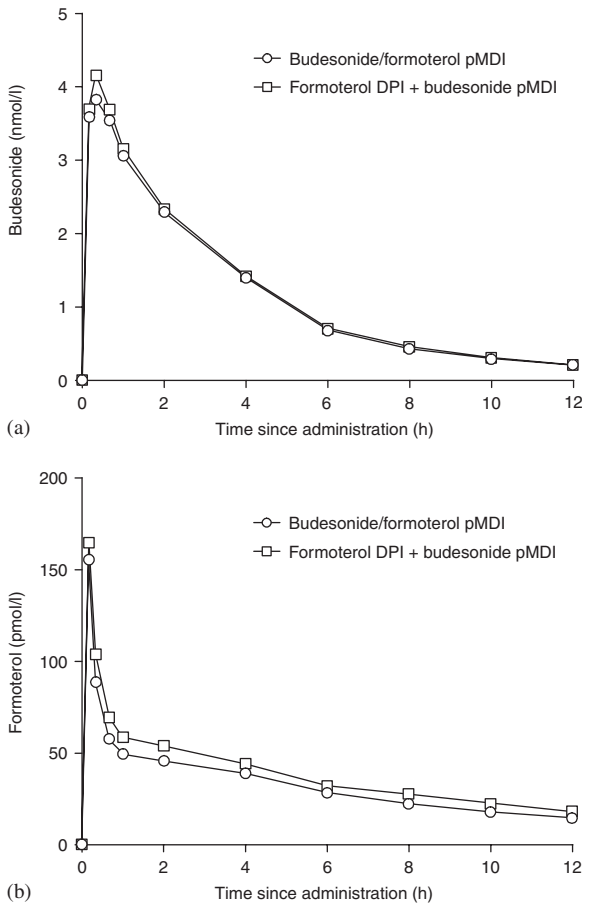

Figure 1. Study II, mean plasma concentration versus time curves for budesonide (a) and formoterol (b) after single-dose administration of budesonide/formoterol pMDI 160/4.5 μg × eight inhalations and budesonide pMDI 160 μg × eight inhalations+formoterol DPI 4.5 μg × eight inhalations. pMDI, pressurized metered-dose inhaler; DPI, dry powder inhaler

nonparametric models. AUC_{0-t} was calculated using the trapezoidal method, with t denoting the last measurement with a concentration above LOQ. $AUC_{0-\infty}$ was calculated as $AUC_{0-t}+C_t/k_{el}$, where C_t is the concentration at time t and k_{el} is the estimated terminal elimination rate constant. k_{el} was calculated from the later plasma concentrations of budesonide and formoterol by performing linear regression on a number of points selected on the nearly linear portion of the ln C(t) versus t curve. The last three points were initially selected and further time points were added as long as the r^2 of the regression model did not decrease. If a smaller r^2 was encountered, the point was excluded and the next point was included in a new regression. If r^2 then was higher, the excluded point was considered an outlier and the process continued; otherwise, the selection process stopped. The late half-life ($t_{1/2}$) was computed as $\ln(2)/k_{el}$.

For repeated-dose treatments in Study IV, $AUC_{0-12\,h}$ was calculated for the interval 0–12 h after the last dose using the trapezoidal method. If the plasma concentration was below LOQ before 12 h, the last part was calculated by exponential extrapolation using k_{el} and the last measurement with concentration above LOQ. The accumulation ratio (R_{ac}) was computed as the ratio of $AUC_{0-12\,h}$ for the repeated dose treatment over the $AUC_{0-12\,h}$ for the single-dose treatment.

In Study II, the amount of formoterol excreted unchanged in the urine (A_e) was calculated as the urine formoterol concentration multiplied by the urine volume. Urine volume was calculated from urine weight, assuming a density of 1020 g/l. The fraction of formoterol excreted in the urine (F_e) was computed from A_e and the inhaled dose (36 μg, corresponding to 86 nmol). The molecular weight of formoterol is 420.4 g/mol.

Safety assessments

Safety and tolerability were based primarily on the incidence and severity of adverse events (AEs) collected using spontaneous subject reports and standard questioning at each clinic visit.

Statistical analysis

The studies were descriptive, and sample sizes were based on pharmacokinetic variability observed in previous studies. Pharmacokinetic analysis in all four studies included all randomized subjects with data collected from at least two treatments with the investigational product. *AUC* values for treatments in each study were compared between treatments using a multiplicative analysis of variance (ANOVA) model with subject, period and treatment as fixed factors. Treatment ratios were estimated and 90% two-sided confidence intervals (CIs) were calculated from the model. C_{max}, $t_{1/2}$ and A_e were analysed using the same methods.

DOI: 10.1002/bdd

Table 4. Comparison of pharmacokinetic parameters[a] for budesonide and formoterol administered via one pMDI versus separate inhalers (study II)

Parameter	BUD/FM pMDI $8 \times 160/4.5\,\mu g$	BUD pMDI $8 \times 160\,\mu g$ +FM DPI $8 \times 4.5\,\mu g$	Treatment comparison
			BUD/FM pMDI[b] vs BUD pMDI[c]+FM DPI[d]
	Mean (90% CI)		Mean ratio (90% CI)
Budesonide			
AUC, nmol.h/l	14.6 (14.0–15.3)	14.9 (14.3–15.6)	97.9 (92.1–104.1)
C_{max}, nmol/l	3.9 (3.7–4.2)	4.0 (3.8–4.3)	97.2 (89.0–106.1)
$t_{1/2}$, h	3.6 (3.4–3.8)	3.3 (3.2–3.5)	107.8 (100.3–115.8)
Formoterol			
AUC, pmol.h/l	515.7 (489.2–543.7)	627.1 (594.8–661.2)	82.2 (76.3–88.6)
C_{max}, pmol/l	147.0 (135.1–160.0)	158.2 (145.4–172.2)	92.9 (82.5–104.7)
$t_{1/2}$, h	6.0 (5.5–6.5)	6.4 (5.9–6.9)	93.8 (83.4–105.4)
Formoterol urine data	Mean (range)		
Ae,[e] nmol	7.6 (4.3–15.0)	9.3 (6.2–14.2)	—
$Ae_{0-24\,h}$, nmol	6.8 (3.8–13.7)	8.3 (5.4–12.9)	—
$Ae_{24-48\,h}$, nmol	0.8 (0.4–1.3)	0.9 (0.5–1.5)	—
Fe,[e] %	8.8 (5.0–17.5)	10.9 (7.3–16.6)	—
$Fe_{0-24\,h}$, %	8.0 (4.4–16.0)	9.7 (6.3–15.0)	—
$Fe_{24-48\,h}$, %	0.9 (0.4–1.5)	1.1 (0.6–1.7)	—

pMDI, pressurized metered-dose inhaler; CI, confidence interval; BUD, budesonide; FM, formoterol; DPI, dry powder inhaler; *AUC*, area under the plasma concentration versus time curve from time zero to infinity; C_{max}, maximum plasma concentration; $t_{1/2}$, elimination half-life; *Ae*, amount excreted unchanged into urine; *Fe*, fraction of the administered dose excreted in the urine.

[a]Values represent geometric means for all parameters; comparisons are shown as percentages based on ratios (90% confidence interval).

[b]Eight inhalations of BUD/FM pMDI 160/4.5 μg.

[c]Eight inhalations of BUD pMDI 160 μg.

[d]Eight inhalations of FM DPI 4.5 μg.

[e]Includes the whole collection period.

In Study III, another model was used to investigate the dose proportionality of budesonide. A multiplicative ANOVA model with subject and period as fixed factors and logged nominal delivered dose as a covariate was applied to budesonide $AUC_{0-\infty}$. The coefficient of the covariate was estimated and 95% CIs were calculated. The estimated slope of the covariate was determined and the regression line was fit to individual and mean values; a covariate slope with 95% CI encompassing 1 indicated dose proportionality.

A post hoc analysis was conducted in Study III to compare the proportionality of budesonide *AUC* with the fine particle dose (FPD). The FPD was determined by Andersen impaction (amount [μg] per inhalation consisting of particles $< 4.7\,\mu m$) of budesonide from the actual batches. The dose-proportionality analysis was not performed on formoterol because the dose was constant and therefore did not change with formulation strength. ANOVA-adjusted estimates of $AUC_{0-12\,h}$ were used to calculate R_{ac} values for budesonide and formoterol.

All randomized subjects who received at least one dose of the investigational product were included in the analysis of safety. AEs were summarized descriptively.

Results

Subjects

In each study, 28 unique subjects were randomized (Table 2), and no subject participated in more than one study. Demographic characteristics of randomized subjects were similar across the four studies: mean age ranged from 24 to 26 years, mean weight from 69 to 72 kg, mean height from 174 to 177 cm, and mean body mass index was about 23 kg/m^2 in all studies (Table 2).

DOI: 10.1002/bdd

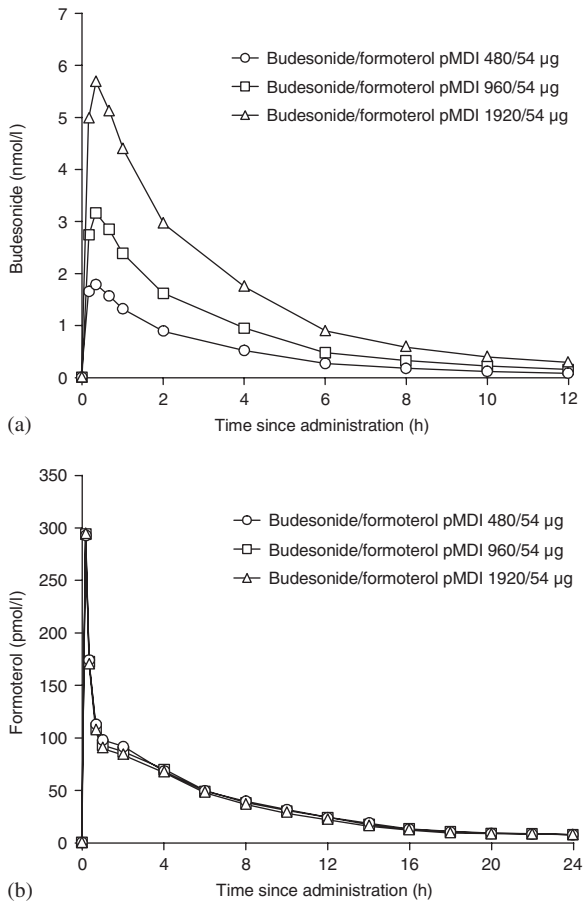

Figure 2. Study III, mean plasma concentration versus time curves for budesonide (a) and formoterol (b) after single-dose administration of 12 inhalations of budesonide/formoterol pMDI at three different doses: 40/4.5 µg, 80/4.5 µg and 160/4.5 µg. pMDI, pressurized metered-dose inhaler

Study I: Pharmacokinetics of budesonide and formoterol administered simultaneously via separate inhalers (budesonide pMDI+formoterol DPI) versus each product alone

Budesonide and formoterol did not show any differences in any pharmacokinetic parameters when administered concomitantly (budesonide pMDI+formoterol DPI) or individually as monocomponents (budesonide pMDI or formoterol DPI) (Table 3). For $AUC_{0-\infty}$ and C_{max}, the 90% CIs for the mean treatment ratios were within the established bioequivalence limits of 80% to 125%.

Study II: Pharmacokinetics of budesonide and formoterol administered from one pMDI inhaler (budesonide/formoterol pMDI) versus separate inhalers (budesonide pMDI+formoterol DPI)

The mean plasma concentration versus time curves for budesonide and formoterol for each treatment are shown in Figure 1. For budesonide, the $AUC_{0-\infty}$ and C_{max} ratios comparing budesonide/formoterol pMDI with budesonide pMDI+formoterol DPI (concomitant administration) were close to 100% (Table 4), with 90% CIs for the mean treatment ratios within the established systemic bioequivalence limits of 80% to 125%. Budesonide $t_{1/2}$ was similar for both treatments. Plasma concentrations of formoterol were slightly lower after treatment with budesonide/formoterol pMDI than after budesonide pMDI+formoterol DPI (concomitant administration). Formoterol $t_{1/2}$ was similar for both treatments.

The estimated relative systemic bioavailability for formoterol, comparing budesonide/formoterol pMDI to budesonide pMDI+formoterol DPI (concomitant administration), was 81.5% based on A_e (in urine), similar to the estimate of relative systemic bioavailability from plasma sampling ($AUC_{0-\infty}$, 82.2%) (Table 4). Values for A_e and F_e are shown in Table 4. The results for $Fe_{0-24\,h}$ and $Fe_{24-48\,h}$ indicate that the majority of formoterol was excreted in the first 24 h.

Study III: Pharmacokinetic dose proportionality of budesonide and bioavailability of formoterol after administration of budesonide/formoterol pMDI at three formulation strengths

Figure 2 shows the mean plasma concentration versus time curves for budesonide and formoterol for each treatment. The dose-adjusted budesonide $AUC_{0-\infty}$ and C_{max} values decreased with increased nominal budesonide dose (Table 5). Twelve inhalations of budesonide/formoterol pMDI 80/4.5 µg resulted in approximately 10% lower values than 12 inhalations of budesonide/formoterol pMDI 40/4.5 µg, and 12 inhalations of budesonide/formoterol pMDI 160/4.5 µg resulted in 10 to 15% lower values than 12 inhalations of budesonide/formoterol pMDI 80/4.5 µg. Budesonide $t_{1/2}$ was similar for the three treatments. Dose proportionality was investigated by determining the estimated slope of the

DOI: 10.1002/bdd

Table 5. Comparisons of pharmacokinetic parameters[a] for the three strengths of budesonide and formoterol administered via one pMDI (study III; budesonide: 12-h data, formoterol: 24-h data)

Parameter	BUD/FM pMDI 12 × 40/4.5 µg	BUD/FM pMDI 12 × 80/4.5 µg	BUD/FM pMDI 12 × 160/4.5 µg	Treatment comparisons,[b] mean ratio (90% CI)		
				BUD/FM pMDI 12 × 40/4.5 µg vs BUD/FM pMDI 12 × 80/4.5 µg	BUD/FM pMDI 12 × 40/4.5 µg vs BUD/FM pMDI 12 × 160/4.5 µg	BUD/FM pMDI 12 × 80/4.5 µg vs BUD/FM pMDI 12 × 160/4.5 µg
Budesonide						
AUC, nmol.h/l	6.0 (5.8–6.3)	10.8 (10.3–11.3)	19.6 (18.7–20.5)	89.6 (84.2–95.3)	81.2 (76.3–86.4)	90.7 (85.2–96.5)
C_{max}, nmol/l	1.8 (1.7–1.9)	3.2 (3.0–3.4)	5.4 (5.0–5.8)	89.7 (81.6–98.6)	76.2 (69.2–83.9)	85.0 (77.2–93.5)
$t_{1/2}$, h	3.6 (3.3–3.8)	3.6 (3.4–3.8)	3.7 (3.5–4.0)	100.5 (91.4–110.4)	104.3 (94.8–114.8)	103.8 (94.4–114.3)
Formoterol						
AUC, pmol.h/l	944.3 (904.5–985.8)	935.3 (896.0–976.5)	909.5 (870.1–950.6)	99.1 (93.2–105.3)	96.3 (90.6–102.4)	97.2 (91.4–103.4)
C_{max}, pmol/l	274.8 (257.5–293.2)	274.9 (257.6–293.2)	271.1 (253.6–289.8)	100.0 (91.3–109.6)	98.7 (89.9–108.3)	98.6 (89.9–108.2)
$t_{1/2}$, h	7.1 (6.3–8.0)	7.8 (7.0–8.8)	8.3 (7.4–9.4)	110.1 (93.3–129.9)	116.6 (98.6–137.9)	105.9 (89.5–125.2)

pMDI, pressurized metered-dose inhaler; CI, confidence interval; BUD, budesonide; FM, formoterol; *AUC*, area under the plasma concentration versus time curve from time zero to infinity; C_{max}, maximum plasma concentration; $t_{1/2}$, elimination half-life.

[a]Values represent geometric means for all parameters; comparisons are shown as percentages based on adjusted mean or ratio (90% confidence interval).

[b]Dose-adjusted *AUC* and C_{max} values are used in treatment comparisons.

covariate, with the regression line fit to individual and mean values; a covariate slope with 95% CI encompassing 1 indicated dose proportionality. The mean slope of the ANOVA covariate (logged nominal delivered dose) was 0.85 (95% CI: 0.80–0.90). Thus, proportionality between budesonide $AUC_{0-\infty}$ and the nominal delivered dose was not demonstrated by this criterion. However, a post hoc analysis of budesonide $AUC_{0-\infty}$ versus the actual FPD was performed for the actual batches used in the study; the budesonide FPD per actuation used in the analysis was 25, 46 and 86 µg for the 40/4.5, 80/4.5 and 160/4.5 µg products, respectively. FPD is related more closely to the amount of drug reaching the lungs than is nominal dose and thus may be a better surrogate parameter for the desired local delivery. The analysis resulted in a mean covariate slope of 0.95 (95% CI: 0.89–1.01), indicating dose proportionality between FPD and plasma levels.

The relative systemic bioavailability of formoterol was comparable when administered via three different formulation strengths of budesonide/formoterol pMDI. The mean treatment comparison ratios for $AUC_{0-\infty}$ and C_{max} had 90% CIs within the established bioequivalence limits of 80 to 125%. The $t_{1/2}$ for formoterol was similar among the three treatments (Table 5).

Study IV: Pharmacokinetic dose proportionality of budesonide and formoterol after administration of budesonide/formoterol pMDI at two different doses for 5 days and pharmacokinetics of repeated versus single dosing

The mean plasma concentration versus time curves for budesonide and formoterol for each treatment are shown in Figure 3, and mean pharmacokinetic parameters and treatment ratios are provided in Table 6. For budesonide, the dose-adjusted $AUC_{0-12\,h}$ and C_{max} ratios comparing four versus two inhalations twice daily for 5 days were close to the expected 100%, with the 90% CI within the bioequivalence limits (80–125%), indicating dose proportionality. Repeated dosing resulted in higher plasma concentrations of budesonide than a corresponding single dose as measured by *AUC* ($AUC_{0-12\,h}$ for repeated dosing and $AUC_{0-\infty}$ for single dosing) and C_{max}.

DOI: 10.1002/bdd

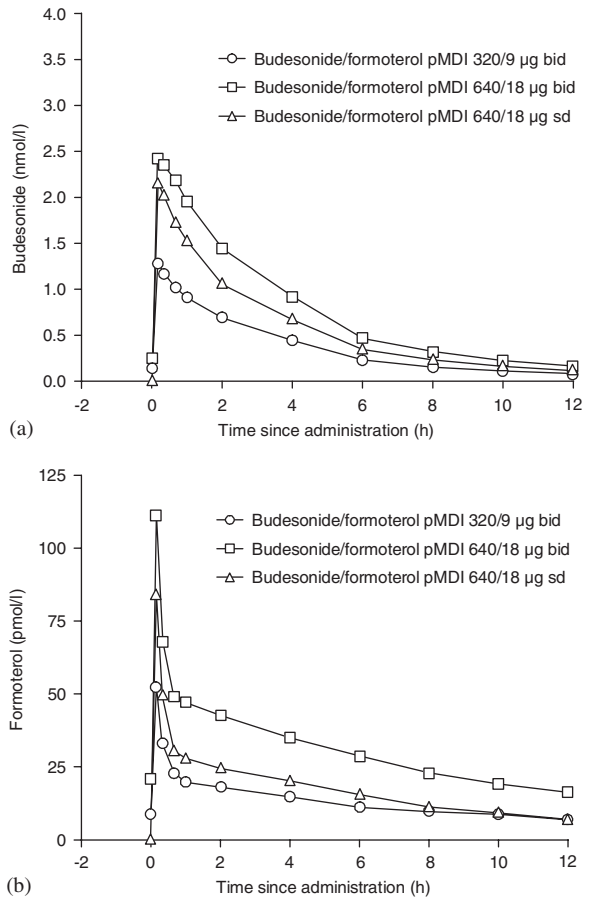

Figure 3. Study IV, mean plasma concentration versus time curves for budesonide (a) and formoterol (b) after 5 days of administration of budesonide/formoterol pMDI 160/4.5 µg × two or four inhalations b.i.d. and single-dose administration of budesonide/formoterol pMDI 160/4.5 µg × four inhalations. pMDI, pressurized metered-dose inhaler; b.i.d., twice daily; s.d., single dose

The R_{ac} for budesonide, computed as the ratio of $AUC_{0–12\,h}$ for repeated dosing (8.8 nmol · h/l) over the $AUC_{0–12\,h}$ for single dosing (6.7 nmol · h/l), was 132.3%. No meaningful differences for budesonide $t_{1/2}$ were found between the two repeated-dosing treatments or between single dosing and repeated dosing (Table 6).

For formoterol, the dose-adjusted $AUC_{0–12\,h}$ and C_{max} ratios, expressed as percentages, comparing four versus two inhalations twice daily for 5 days were higher than the expected 100% (Table 6). No meaningful differences were found between the two repeated-dosing treatments for formoterol $t_{1/2}$. Repeated dosing resulted in higher plasma concentrations of formoterol than a corresponding single dose as measured by *AUC* ($AUC_{0–12\,h}$ for repeated dosing and $AUC_{0–\infty}$ for single dosing) and C_{max}. The R_{ac} for formoterol, calculated as the ratio of $AUC_{0–12\,h}$ for repeated dosing (356.6 pmol · h/l) over the $AUC_{0–12\,h}$ for single dosing (201.0 pmol · h/l), was 177.4%. The higher formoterol plasma concentrations observed with repeated-dose treatment compared with single-dose treatment also was reflected in a relatively longer $t_{1/2}$ after repeated dosing (Table 6).

Safety

All treatments were well tolerated, with no new or unexpected safety findings in these studies. The most common AEs were tremor, headache and palpitations, which are well known and consistent with high-dose β_2-adrenergic agonist administration [21]. Tremor and palpitations occurred more frequently in subjects who received higher or more frequent doses of formoterol compared with lower or less frequent doses in Studies I and IV. In addition, the occurrence of tremor and palpitations was more frequent when the same dose of formoterol was administered via DPI compared with budesonide/formoterol pMDI in Study II. Nasopharyngitis also was common, possibly because the studies were performed during winter. There were no clinically relevant differences in the pattern of reported AEs between treatments.

Discussion

When pharmaceutical agents with known efficacy and tolerability are combined in a new formulation, it is necessary to investigate possible interactions of the components. Systemic absorption of corticosteroids has the potential to produce adverse effects such as hypothalamic–pituitary–adrenal axis suppression, slowing of growth in children and osteoporosis in older individuals [22,23]. Systemic exposure to β_2-adrenergic receptor agonists has been associated with AEs, including headache, tremor, muscle

DOI: 10.1002/bdd

Table 6. Comparisons of pharmacokinetic parameters[a] for the two doses of budesonide and formoterol administered via one pMDI and using repeated versus single-day dosing (study IV)

Parameter[a]	Repeated dosing		Single-dose	Treatment comparisons,[b] mean ratio (90% CI)	
	BUD/FM pMDI $2 \times 160/4.5\,\mu g$ b.i.d.	BUD/FM pMDI $4 \times 160/4.5\,\mu g$ b.i.d.	BUD/FM pMDI $4 \times 160/4.5\,\mu g$ s.d.	BUD/FM pMDI $4 \times 160/4.5\,\mu g$ b.i.d. vs BUD/FM pMDI $2 \times 160/4.5\,\mu g$ b.i.d.	BUD/FM pMDI $4 \times 160/4.5\,\mu g$ b.i.d. vs BUD/FM pMDI $4 \times 160/4.5\,\mu g$ s.d.
Budesonide					
AUC,[c] nmol.h/l	4.2 (4.0–4.5)	8.8 (8.3–9.3)	7.3 (6.9–7.7)	104.3 (96.6–112.6)	121.2 (112.2–130.8)
C_{max}, nmol/l	1.2 (1.1–1.3)	2.4 (2.2–2.6)	2.1 (1.9–2.3)	100.4 (90.0–112.0)	115.6 (103.6–128.9)
$t_{1/2}$, h	4.0 (3.8–4.3)	3.8 (3.5–4.0)	3.7 (3.5–4.0)	93.6 (85.5–102.4)	100.9 (92.2–110.4)
Formoterol					
AUC,[c] pmol.h/l	151.7 (145.2–158.4)	356.7 (341.4–372.6)	256.4 (245.4–267.8)	117.6 (110.5–125.1)	139.1 (130.8–148.0)
C_{max}, pmol/l	47.7 (44.5–51.1)	104.6 (97.6–112.1)	77.0 (71.8–82.5)	109.7 (99.4–121.0)	135.8 (123.1–149.8)
$t_{1/2}$, h	6.9 (6.3–7.7)	7.0 (6.3–7.7)	5.3 (4.8–5.8)	100.0 (87.0–115.0)	131.2 (114.2–150.8)

pMDI, pressurized metered-dose inhaler; CI, confidence interval; BUD, budesonide; FM, formoterol; b.i.d., twice daily; s.d., single dose; *AUC*, area under the plasma concentration versus time curve; C_{max}, maximum plasma concentration; $t_{1/2}$, elimination half-life.

[a]Values represent geometric means for all parameters; comparisons are shown as percentages based on adjusted mean or ratio (90% confidence interval).

[b]Dose-adjusted *AUC* and C_{max} values are used in treatment comparisons.

[c]0–12 h for b.i.d. dosing, 0–∞ for single dosing.

cramps, palpitations and decreased serum potassium [21]. In both cases, these concerns are reduced by the favorable pharmacokinetics achieved with inhalation therapy, which delivers medication directly to the lungs with minimal systemic exposure [24,25]. Both budesonide and formoterol, delivered separately, are efficacious and have acceptable tolerability [26–28]. The present studies, the first to explore the pharmacokinetics of budesonide and formoterol administered together in one pMDI in healthy adults, are critical in determining the systemic exposure, one important determinant for the safety profile of this combination product.

The similarity of pharmacokinetic parameters for budesonide and formoterol administered simultaneously with separate inhalers and as individual monocomponents demonstrated in Study I indicates that there is no pharmacokinetic interaction between budesonide and formoterol. These results also suggest consistent dose delivery from the investigated drug formulations on the different experimental days. The lack of pharmacokinetic interactions between budesonide and formoterol is in line with previous studies of budesonide/formoterol DPI [20]. Our findings complement those reported by Cazzola and colleagues, in which improvements in lung function were observed with formoterol/budesonide DPI compared with formoterol DPI alone without a concomitant modification of heart rate, indicating a lack of interaction of formoterol and budesonide translating to a systemic effect [29]. A lack of pharmacokinetic interaction also has been observed with another corticosteroid/LABA combination, salmeterol and fluticasone, when administered using a DPI [30].

Study II demonstrated that systemic exposure to budesonide from the combination pMDI was nearly identical to that from the budesonide pMDI, which had the same formulation with the exception of formoterol. Systemic exposure (*AUC*) to formoterol from the combination pMDI was about 20% lower than that from the formoterol DPI. In contrast, Houghton and colleagues reported similar systemic absorption of formoterol with both pMDI and DPI administration, resulting in similar systemic effects [31].

The estimate of the late elimination half-life for formoterol in Studies I and II ($\sim$6 h) was shorter than those observed in Study III ($\sim$8 h), which was based on a longer 24-h plasma sampling. While plasma concentrations were quantifiable for the full 12-h sampling period, the interval may have been too short to obtain an accurate estimate of the late elimination half-life. An underestimated late elimination half-life and large extrapolated area also might be expected

DOI: 10.1002/bdd

to result in a lower estimate of total *AUC*. However, treatments in each study had similar $AUC_{0-\infty}$ values, implying that any systematic underestimation was uniform. The treatment ratios for AUC_{0-t} (data not shown) and C_{max}, both based on actual measurements without extrapolation, were very similar to the treatment ratios for $AUC_{0-\infty}$, suggesting that the influence of any underestimation was probably small. Finally, formoterol data from two different methods in Study II (48-h urine sampling and 12-h plasma sampling) were similar, suggesting that the plasma-based estimates of formoterol bioavailability were valid.

In Study III, the pharmacokinetic parameters for formoterol were calculated using measurements from 0 to 12 h (data not shown), as well as from 0 to 24 h, to compare the effect of sampling time on the pharmacokinetic parameters. The results show that the *AUC* values, whether calculated from values up to 12-h or up to 24-h, were comparable for all budesonide/formoterol pMDI formulation strengths, with 90% CIs within the established bioequivalence limits. However, as expected, the estimate of the late elimination half-life was shorter for all treatments using the 0 to 12 h data (~5 h) compared with the 0 to 24-h data (~8 h). This also is consistent with results of Studies I, II and IV, in which values for the formoterol late elimination half-life derived from 12-h plasma sampling were all lower than that derived from 24-h plasma sampling in Study III. Although a compartmental analysis was not performed in the present studies, two elimination phases for formoterol metabolites were observed in a study by Rosenborg *et al*., which evaluated the metabolism of formoterol after combined intravenous and oral administration [32].

Pharmacokinetic dose proportionality after treatment with budesonide/formoterol pMDI was examined in Studies III and IV. In Study III, the formulation strength of budesonide was varied, while the formoterol dose was kept constant. For budesonide, the 90% CI for the treatment comparison ratios of the dose-adjusted $AUC_{0-\infty}$ were within the established bioequivalence limits for adjacent doses (12 inhalations of 40/4.5 μg vs 80/4.5 μg or 12 inhalations of 80/4.5 μg vs 160/4.5 μg), indicating that the plasma concentration of budesonide changed in proportion with the nominal delivered dose for two adjacent formulation strengths. A more thorough assessment of proportionality across the three treatments demonstrated that $AUC_{0-\infty}$ increased in proportion with FPD, but slightly less than proportionally with the nominal delivered dose. However, the fine particle dose may be more appropriate than the nominal dose in assessing dose proportionality, as it is more representative of the dose delivered to the lungs [33] and because most of the drug that is swallowed undergoes first-pass metabolism and is not absorbed into the systemic circulation [34]. The $AUC_{0-\infty}$ of formoterol was similar across the three budesonide formulation strengths, with 90% CIs of the mean treatment comparison ratios for $AUC_{0-\infty}$ and C_{max} within established bioequivalence limits. These data indicate that switching between formulation strengths with varying doses of budesonide has no effect on the plasma concentration of formoterol.

In Study IV, comparing twice-daily dosing over 5 days with four inhalations of budesonide/formoterol pMDI 160/4.5 μg versus two inhalations of budesonide/formoterol pMDI 160/4.5 μg, the dose-adjusted $AUC_{0-12\,h}$ and C_{max} ratios for budesonide were within the bioequivalence limits, indicating proportionality. These data demonstrate that an increase in dose of budesonide/formoterol pMDI from two to four inhalations would coincide with an increase in systemic exposure to budesonide proportional to the dose. These results are consistent with those of Kaiser *et al.* who demonstrated that plasma budesonide concentrations were proportional to the administered dose after single and multiple doses of budesonide DPI 400 μg twice daily, 800 μg twice daily and 1600 μg twice daily in adults with mild asthma [14]. For formoterol in Study IV, the 90% CI for the dose-adjusted $AUC_{0-12\,h}$ ratios (four vs two inhalations) fell just outside the bioequivalence limits (125.1%), but C_{max} fell within the bioequivalence limits. The difference in dose-adjusted $AUC_{0-12\,h}$ may be due to plasma concentrations falling below the LOQ for some subjects before 12 h with the low-dose treatment, probably resulting in a slight underestimation of $AUC_{0-12\,h}$ relative to the high-dose treatment.

DOI: 10.1002/bdd

Study IV also included a comparison of single and repeated dosing with budesonide/formoterol pMDI. Increased systemic exposure with repeated dosing in Study IV was also reflected in R_{ac}, which was 132% for budesonide and 177% for formoterol. The results with budesonide are consistent with previous findings [14], and the accumulation ratio for formoterol is similar to that reported for patients with asthma (163% to 208%) based on urinary formoterol excretion observed with another marketed version of inhaled formoterol (Foradil® Aerolizer®, Novartis Pharma AG, Basel, Switzerland) [35].

The pharmacokinetic data for budesonide/formoterol pMDI suggest no additional safety concerns over that observed when the individual components are administered concomitantly via separate inhalers or alone.

In summary, the four studies presented here demonstrate that there is no pharmacokinetic interaction between budesonide and formoterol when delivered concomitantly via two inhalers or individually as monocomponents. In addition, systemic exposure to budesonide from the combination pMDI was similar to that from budesonide pMDI, and the systemic exposure after an increase in budesonide dose was well correlated to the increases in fine particle dose. Systemic exposure to formoterol from the combination pMDI was about 20% lower than that from the formoterol DPI. The studies also indicate comparable bioavailability of formoterol when administered via three different formulation strengths of budesonide/formoterol pMDI. Finally, the studies suggest that an increase in dose of budesonide/formoterol pMDI from two to four inhalations would coincide with an increase in systemic exposure proportional to the dose.

Acknowledgements

It is a pleasure to acknowledge Dr Jonas Pettersson and the staff at the AstraZeneca Clinical Pharmacology Unit, Lund, Sweden, for the performance of the studies, as well as Lisa M. Klumpp, PhD, and Cynthia Gobbel, PhD, from Scientific Connexions (Newtown, PA, USA) for assistance in writing the manuscript. Editorial support was funded by AstraZeneca LP. All authors are employees of AstraZeneca. This study was sponsored by AstraZeneca.

References

1. National Asthma Education and Prevention Program. Expert panel report: guidelines for the diagnosis and management of asthma. Update on selected topics – 2002. *J Allergy Clin Immunol* 2002; **110**: S141–S219.
2. Global Initiative for Asthma (GINA). Global Strategy for Asthma Management and Prevention. 2006 Update. Available at: http://www.ginasthma.com/Guidelineitem.asp?l1=2&l2=1&intId=60.
3. Global Initiative for Chronic Obstructive Lung Disease. Global Strategy for the Diagnosis, Management, and Prevention of Chronic Obstructive Pulmonary Disease. 2005 Update. Available at: http://www.goldcopd.com/download.asp?intID=234.
4. Stoloff SW, Stempel DA, Meyer J, Stanford JH, Carranza Rosenzweig JR. Improved refill persistence with fluticasone propionate and salmeterol in a single inhaler compared with other controller therapies. *J Allergy Clin Immunol* 2004; **113**: 245–251. doi:10.1016/j.jaci.2003.10.011
5. Bender B, Milgrom H, Rand C. Nonadherence in asthmatic patients: is there a solution to the problem? *Ann Allergy Asthma Immunol* 1997; **79**: 177–186.
6. Buhl R, Creemers JPHM, Vondra V, Martelli NA, Naya IP, Ekström T. Once-daily budesonide/formoterol in a single inhaler in adults with moderate persistent asthma. *Respir Med* 2003; **97**: 323–330.
7. Lalloo UG, Malolepszy J, Kozma D, *et al*. Budesonide and formoterol in a single inhaler improves asthma control compared with increasing the dose of corticosteroid in adults with mild-to-moderate asthma. *Chest* 2003; **123**: 1480–1487. doi:10.1378/chest.123.5.1480
8. Tal A, Simon G, Vermeulen JH, *et al*. Budesonide/formoterol in a single inhaler versus inhaled corticosteroids alone in the treatment of asthma. *Pediatr Pulmonol* 2002; **34**: 342–350. doi:10.1002/ppul.10173
9. Zetterström O, Buhl R, Mellem H, *et al*. Improved asthma control with budesonide/formoterol in a single inhaler, compared with budesonide alone. *Eur Respir J* 2001; **18**: 262–268.
10. Ankerst J, Persson G, Weibull E. Tolerability of a high dose of budesonide/formoterol in a single inhaler in patients with asthma. *Pulm Pharmacol Ther* 2003; **16**: 147–151.
11. Calverley PM, Boonsawat W, Cseke Z, Zhong N, Peterson S, Olsson H. Maintenance therapy with budesonide and formoterol in chronic obstructive pulmonary disease. *Eur Respir J* 2003; **22**: 912–919.
12. Szafranski W, Cukier A, Ramirez A, *et al*. Efficacy and safety of budesonide/formoterol in the management of chronic obstructive pulmonary disease. *Eur Respir J* 2003; **21**: 74–81.

13. Donnelly R, Seale JP. Clinical pharmacokinetics of inhaled budesonide. *Clin Pharmacokinet* 2001; **40**: 427–440. doi:10.2165/00003088-200140060-00004
14. Kaiser H, Aaronson D, Dockhorn R, Edsbäcker S, Korenblat P, Källén A. Dose-proportional pharmacokinetics of budesonide inhaled via Turbuhaler®. *Br J Clin Pharmacol* 1999; **48**: 309–316. doi:10.1046/j.1365-2125.1999.00008.x
15. Minto C, Li B, Tattam B, Brown K, Seale JP, Donnelly R. Pharmacokinetics of epimeric budesonide and fluticasone propionate after repeat dose inhalation—intersubject variability in systemic absorption from the lung. *Br J Clin Pharmacol* 2000; **50**: 116–124. doi:10.1046/j.1365-2125.2000.00218.x
16. Agertoft L, Andersen A, Weibull E, Pedersen S. Systemic availability and pharmacokinetics of nebulised budesonide in preschool children. *Arch Dis Child* 1999; **80**: 241–247.
17. Lecaillon JB, Kaiser G, Palmisano M, Morgan J, Della Cioppa G. Pharmacokinetics and tolerability of formoterol in healthy volunteers after a single high dose of Foradil dry powder inhalation via Aerolizer™. *Eur J Clin Pharmacol* 1999; **55**: 131–138. doi:10.1007/s002280050607
18. Bartow RA, Brogden RN. Formoterol. An update of its pharmacological properties and therapeutic efficacy in the management of asthma. *Drugs* 1998; **55**: 303–322. doi:10.2165/00003495-199855020-00016
19. Thorsson L, Edsbäcker S, Källén A, Löfdahl C-G. Pharmacokinetics and systemic activity of fluticasone via Diskus® and pMDI, and of budesonide via Turbuhaler®. *Br J Clin Pharmacol* 2001; **52**: 529–538. doi:10.1046/j.0306-5251.2001.01493.x
20. Goldsmith DR, Keating GM. Budesonide/formoterol: a review of its use in asthma. *Drugs* 2004; **64**: 1597–1618. doi:10.2165/00003495-200464140-00006
21. Sovani MP, Whale CI, Tattersfield AE. A benefit-risk assessment of inhaled long-acting β_2-agonists in the management of obstructive pulmonary disease. *Drug Safety* 2004; **27**: 689–715. doi:10.2165/00002018-200427100-00001
22. Gulliver T, Eid N. Effects of glucocorticoids on the hypothalamic–pituitary–adrenal axis in children and adults. *Immunol Allergy Clin North Am* 2005; **25**: 541–555, vii. doi:10.1016/j.iac.2005.04.001
23. Passalacqua G, Albano M, Canonica GW, *et al*. Inhaled and nasal corticosteroids: safety aspects. *Allergy* 2000; **55**: 16–33. doi:10.1034/j.1398-9995.2000.00370.x
24. Newman SP. Deposition and effects of inhaled corticosteroids. *Clin Pharmacokinet* 2003; **42**: 529–544. doi:10.2165/00003088-200342060-00003
25. Rau JL. The inhalation of drugs: advantages and problems. *Respir Care* 2005; **50:** 367–382.
26. Banov CH. The role of budesonide in adults and children with mild-to-moderate persistent asthma. *J Asthma* 2004; **41**: 5–17. doi:10.1081/JAS-120026092
27. Brogden RN, McTavish D. Budesonide. An updated review of its pharmacological properties, and therapeutic efficacy in asthma and rhinitis. *Drugs* 1992; **44**: 375–407.
28. Berger WE. The use of inhaled formoterol in the treatment of asthma. *Ann Allergy Asthma Immunol* 2006; **97**: 24–33.
29. Cazzola M, Santus P, Di Marco F, *et al*. Onset of action of formoterol/budesonide in single inhaler vs formoterol in patients with COPD. *Pulm Pharmacol Ther* 2004; **17**: 121–125.
30. Kirby S, Falcoz C, Daniel MJ, *et al*. Salmeterol and fluticasone propionate given as a combination. Lack of systemic pharmacodynamic and pharmacokinetic interactions. *Eur J Clin Pharmacol* 2001; **56**: 781–791. doi:10.1007/s002280000233
31. Houghton CM, Langley SJ, Singh SD, *et al*. Comparison of bronchoprotective and bronchodilator effects of a single dose of formoterol delivered by hydrofluoroalkane and chlorofluorocarbon aerosols and dry powder in a double blind, placebo-controlled, crossover study. *Br J Clin Pharmacol* 2004; **58**: 359–366. doi:10.1111/j.1365-2125.2004.02172.x
32. Rosenborg J, Larsson P, Tegnér K, Hallström G. Mass balance and metabolism of [^{3}H]formoterol in healthy men after combined i.v. and oral administration—mimicking inhalation. *Drug Metab Dispos* 1999; **27**: 1104–1116.
33. Borgström L. *In vitro*, *ex vivo*, *in vivo veritas. Allergy* 1999; **54**: 88–92.
34. Tattersfield AE, Harrison TW, Hubbard RB, Mortimer K. Safety of inhaled corticosteroids. *Proc Am Thorac Soc* 2004; **1**: 171–175. doi:10.1513/pats.200402-016MS
35. *Foradil® Aerolizer® (Formoterol Fumarate Inhalation Powder) Prescribing Information*. Novartis Pharma AG: Basel, Switzerland, October 2002.